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(54) Method of producing soy protein hydrolyzate from fat-containing soy material, and such soy protein hydrolyzate

(57) There is provided a method of producing soy protein hydrolyzate from fat-containing soy material (as defined), which method comprises hydrolyzing a partially defatted solid soy material, obtained by washing a fat-containing soy material in an aqueous medium at a pH in the range of from 3.5 to 5.5 at a relatively constant pH with a proteolytic enzyme in the presence of water and a base to a pH in the range of from 1 to 20 and thereafter deactivating the enzyme, whereafter the aqueous hydrolyzate phase is separated from the oil phase and the solid phase, as well as a product produced by such method.

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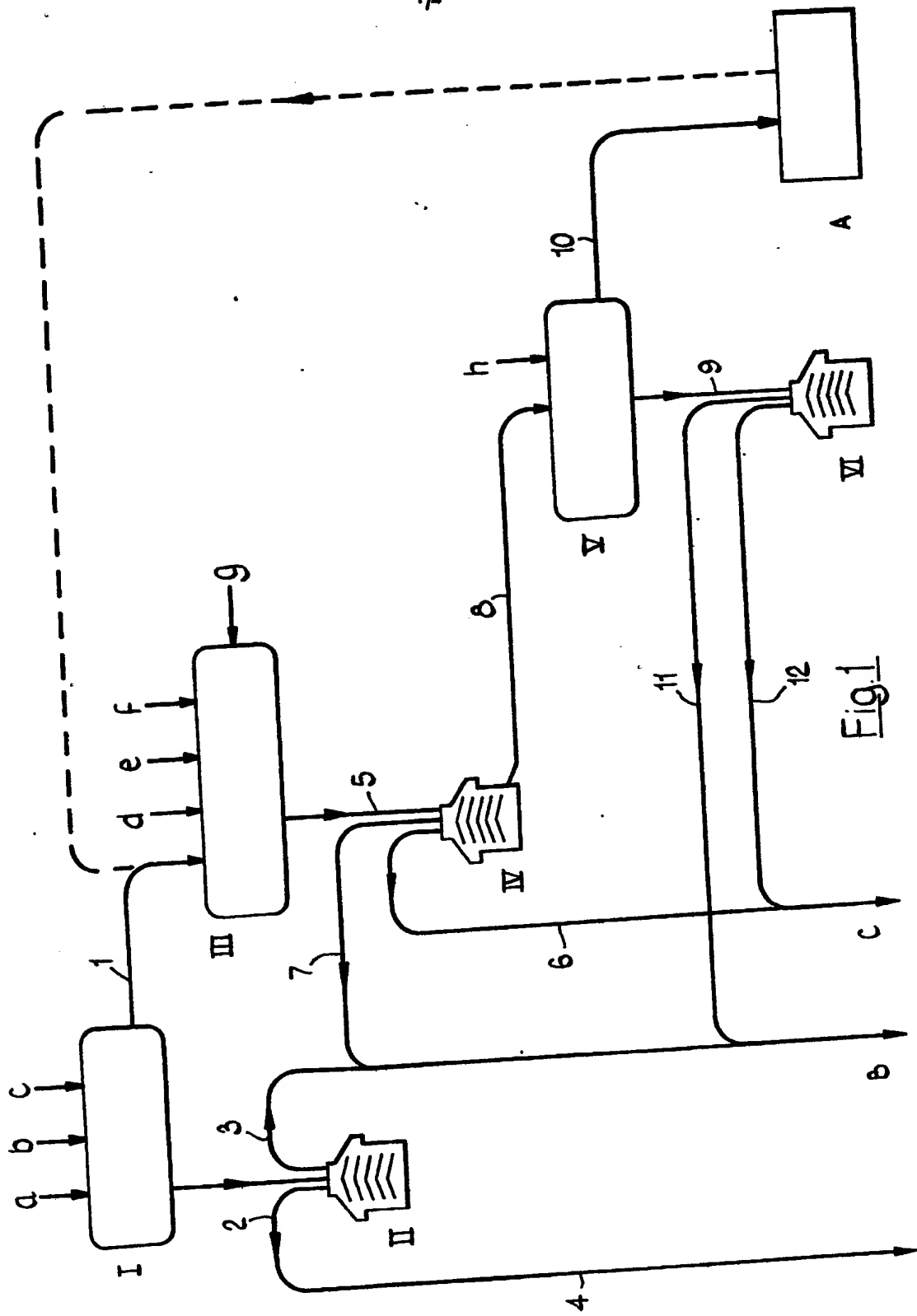


Fig. 1

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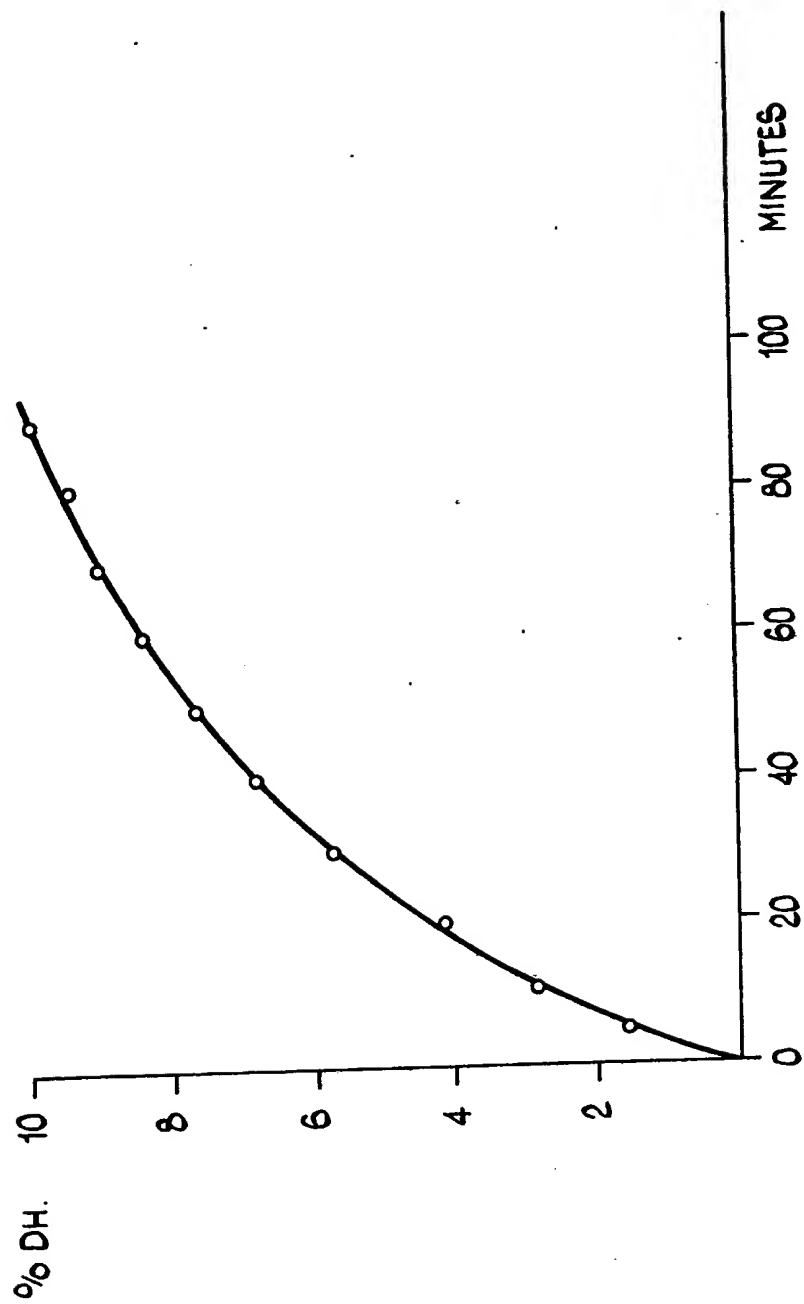


Fig. 2

SPECIFICATION

Method of producing soy protein hydrolyzate from fat-containing soy material, and such soy protein hydrolyzate

- 5 The invention comprises a method of producing soy protein hydrolyzate from fat-containing soy material, and soy protein hydrolyzate so prepared.
- A method for production of soy protein hydrolyzate from soy beans which are defatted by extraction with organic solvents is described for example in Fifth International Congress of Food Science & Technology, 10 Abstract of paper 3b - 14, "Enzymatic hydrolysis of soy protein. Processing developments and applications in low pH foods". However, due to the presence of fat in the fat-containing soy materials used as a starting material in the present invention and the decisive role which is taken by this fat in all processes, in which fat is present, the invention differs radically from the above indicated production of soy protein hydrolyzate from defatted soy beans.
- 15 Soy protein hydrolyzate is a material of growing importance for example for the food industry. Thus, it can be used as one of the main constituent in brines for meat pumping in order to enrich the protein content thereof, as a constituent in soy milk in order to enrich the soy milk with protein without increasing the beany taste normally present in soy milk based on non-hydrolyzed soy material and as a protein enriching agent used as an additive for both acid and neutral soft drinks.
- 20 Herein and in the accompanying claims, the term "fat-containing soy material" is used generically to include full-fat soy flour, ground whole soy beans, crushed soy beans, which are partially defatted by mechanical means and similar materials.
- Fat-containing soy material, especially full-fat soy flour, is available in huge amounts in areas of the world with industry of a primitive nature.
- 25 In any production of a refined protein product with fat-containing soy materials as a starting material, the concomitant fat recovery is important. Usually the recovery of soy oil from soy beans comprises an extraction with organic solvents, generally a hexane extraction. The solvent extraction requires a solvent recovery by fractional distillation, which requires a relatively high investment, and furthermore this process is not ideal from an environmental point of view, especially since ordinarily highly inflammable solvents are 30 used for the extraction. Also, the process is so elaborate that it is not well suited for use at production sites of a primitive nature, for example in developing countries.
- Thus, a need exists for a method for treatment of a fat-containing soy material which is well suited for production sites of a primitive nature and which furthermore gives rise to an organoleptically acceptable soy protein hydrolyzate and a considerable recovery of the soy oil and other valuable materials in the full-fat soy 35 flour.
- The method for production of soy protein hydrolyzate from fat-containing soy material according to the invention comprises hydrolyzing a partially defatted solid soy material, obtained by washing a fat-containing soy material in an aqueous medium at a pH in the range of from 3.5 to 5.5, at a relatively constant pH with a proteolytic enzyme in the presence of water and a base to a DH in the range of from 1 to 20 and thereafter 40 deactivating the enzyme, whereafter the aqueous hydrolyzate phase is separated from the oil phase and the solid phase.
- A preferred embodiment of the method according to the invention includes the step of washing fat-containing soy material, in an aqueous medium having a pH in the range of from 3.5 to 5.5, preferably 4.2 to 4.5.
- 45 Advantageously, the method of producing soy protein hydrolyzate from fat-containing soy material according to the invention comprises washing the fat-containing soy material (a) in an aqueous medium at a pH in the range of from 4.2 to 4.5 (operation I), the wash water (2) from operation I is introduced into a separator, wherein it is separated into an oil phase (3) and a waste water phase (4) (operation II), the washed, partially defatted solid soy material (1) from operation I is introduced into a hydrolysis container, to which 50 also water (d), a proteolytic enzyme (e) and base (f) is added, in which hydrolysis container the partially defatted soy material (1) from operation I is hydrolyzed at a relatively constant pH to a degree of hydrolysis (DH) in the range of from 1 to 20 (operation III), whereafter the proteolytic activity is inactivated, the slurry (5) from operation III is introduced into a separator, in which the slurry is separated into an oil phase (7), an aqueous hydrolyzate phase (6) and a sludge phase (8) (operation IV), the sludge phase (8) from operation IV is 55 collected (product A), the oil phase (3) and (7) from operations II and IV are combined (product B) and the aqueous hydrolyzate phase (6) from operation IV is collected (product C).
- The invention also relates to the hydrolyzates produced by the method of the invention.
- Surprisingly, it is found that it is possible, according to the invention, by means of a method which is well suited for production sites of a primitive nature to recover in a good yield a valuable soy protein hydrolyzate 60 without bitterness, without soy flavour and without any disadvantageous properties originating from the soy fat which has several application possibilities, around 60% of the oil as a separate oil phase and the precipitate from the hydrolysis, which can be used either as a high grade fodder or as a new starting material in the hydrolysis step.
- Surprisingly, it has been found that the soy protein hydrolyzate of the invention can be fully acceptable 65 from an organoleptic point of view and also that the oil phase does not turn rancid during the recovery.

A preferred embodiment of the method according to the invention comprises transporting the sludge phase ⑧ from operation IV before collection to a washing device, to which also water (h) is added (operation VI), whereafter the precipitate ⑩ from operation V is collected as product A, the wash water phase ⑨ from operation V is introduced into a separator, in which it is separated into an oil phase ⑪ and an aqueous hydrolyzate phase ⑫ (operation VI), the oil phases ③, ⑦ and ⑪ from operation II, IV and VI are combined (product B) and the aqueous hydrolyzate phases ⑥ and ⑫ from operations IV and VI are combined (product C). In this way, the content of low molecular compounds, for example low molecular peptides, is washed out from the solid phase (8) from operation IV, and the product A will be well suited for repeated hydrolysis. If too many low molecular peptides are present in the material which is subjected to hydrolysis, in the first place these will be decomposed enzymatically to yield a bitter tasting product, and in the second place the proteolytic enzymes will not primarily - as intended - decompose the high molecular soy protein, but rather primarily decompose the low molecular peptides.

In a preferred embodiment of the method according to the invention, the separations in one or more or all of operations II, IV and VI are performed by means of centrifuges. In this way, a fast and efficient separation is obtained.

In a preferred embodiment of the method according to the invention, the proteolytic enzyme used for the hydrolysis is produced by means of *Bacillus licheniformis*, and that the hydrolysis is performed around the pH optimum of this enzyme. A preferred example of such proteolytic enzyme is the commercial product sold under the Trade Mark "ALCALASE" (subtilisin Carlsberg) by NOVO INDUSTRI A/S. This enzyme is able to split protein along the protein chain with such high hydrolysis rate that the minimal value DH is quickly reached.

It is preferred that the hydrolysis is performed at a pH which does not differ more than 2.5 pH units from the optimum pH of the proteolytic enzyme. The optimum pH of the proteolytic enzyme should be determined by means of a substrate related to the hydrolysis mixture. If for example "ALCALASE" is used as the proteolytic enzyme, the enzyme activity curve and thus the optimal pH activity can be determined by means of the modified Anson method described in NOVO Enzyme Information 1B no. 058 e-GB (the original Anson method is described in J. Gen. Physiol., 22, 79-89 (1939)). According to this method, the pH optimum for "ALCALASE" in the hydrolysis mixture is around 9.0 and the pH during hydrolysis should accordingly in this preferred embodiment of the invention have a value in the range of from 6.5 to 11.5.

In a preferred embodiment of the method according to the invention, the hydrolysis is carried out to a DH in the range of from 8 to 12.

The proteolytic activity is preferably inactivated by means of malic or citric acid.

The hydrolysis may be performed in any desired manner, such as that known *per se*, from the disclosure of United States Patent Specification No. 4,100,024.

Also, the soy oil phase can be purified in any desired manner, for instance by the known *per se* method of removing residual amounts of protein and water.

The degree of hydrolysis (DH) is defined by the equation

$$DH = \frac{\text{Number of peptide bonds cleaved}}{\text{Total number of peptide bonds}} \times 100\%$$

Reference is made to J. Adler-Nissen, J. Agric. Food Chem. Vol. 24 No. 6, (1976) page 1090 - 1093, where a more detailed discussion of the definition of DH appears.

The number of the peptide bonds cleaved can be measured by means of the ninhydrin method. The ninhydrin method is described in Moore, S., Stein, W.H., "Photometric Ninhydrin Method for use in the Chromatography of Amino Acids", J. Biol. Chem., 176, 367-388 (1948).

The DH can also be determined if the course of hydrolysis is followed by means of the pH-STAT method, as described in Jacobsen, S.F., Léonis, J., Linderstrøm-Lang, K., Ottesen, M., "the pH-STAT and its use in Biochemistry", in Glick, D. (edit.), "Methods of Biochemical Analysis", Vol. IV. pp. 171-210, Interscience, Publishers Inc., New York (1957).

It appears from the above that the DH plays an important role in the invention, in as much as the hydrolysis is controlled by means of the DH: only when DH has reached a critical value, the hydrolysis may be terminated. The DH is, so to speak, the main parameter of the hydrolysis.

For a better understanding of the present invention and to show how the same may be put into effect, reference will now be made, by way of example, to the accompanying drawings, in which Figure 1 shows a flow sheet of a preferred embodiment of the method according to the invention, and Figure 2 shows the time - DH relationship pertaining to Example 1.

Referring now to Figure 1, the fat-containing soy material (a), which should be pretreated without formation of off-flavour, is washed (extracted) with water (b) (Operation I). Acid (c) is introduced initially until the pH is in the range of from 4 to 4.5 in the wet soy material, but not later on, because pH turns out to be constant, even if large amounts of water are used. The soy material is washed until it has a bland taste, and until all soluble materials (at pH 4 to 4.5) are removed. A stepwise operation in which each step includes a separation of the liquid and the solid phase may be used, and if a liquid/solid ratio of 10 : 1 is used, operation I can be carried out by means of decanter centrifuges or other types of separators. In this case, at least four

steps are found necessary. Other types of extraction equipment may be used, for example batch centrifuges, continuous or batch-operating counter current extractors or press-equipment. By operation I, a partially defatted and washed soy material (1) is removed. Furthermore, the total amount of wash liquid (2) is recovered and, during operation II, this liquid is separated into an oil-phase (3) and an oil free phase (4), which may be regarded as waste water.

The partially defatted and washed soy material (1) is transferred to a hydrolysis tank equipped with a stirrer, thermometer and pH-electrodes connected to a titrator, in which hydrolysis (operation III) takes place. Water (4) is added to the soy material (1) until the protein concentration is in the range of from 6 to 10% (N x 6.25). The temperature is adjusted to 50 - 55°C and Alcalase is added (e). If the hydrolyzate is intended for nutritional purposes, a food grade preparation of the proteolytic enzyme is used in such amounts that the total hydrolysis time is around two hours.

The enzymatic hydrolysis (operation III) is carried out at constant pH, preferably at pH 8.0. In order to maintain the chosen pH for the reaction, continuous addition of base (f) is necessary during the reaction. As described in Adler-Nissen Process Biochem. 12(6)18, (1977), the DH can be calculated from the consumption of base (f).

When DH reaches the predetermined value, preferably 10%, the hydrolysis is terminated by addition of acid (g) until the pH is 4.0. The enzyme is inactivated after 30 minutes at pH 4.0 and 50°C. When malic or citric acids are used, the hydrolyzate is non bitter; other acids may be used provided they do not interfere disadvantageously with the product to which the hydrolyzate is supposed to be added.

The finished hydrolyzate (6) is then separated (operation IV) into an oil phase (7), a soy protein hydrolyzate (8) and a sludge phase (9), containing insoluble protein, polysaccharides and residual amounts of oil. Preferably, a three-phase-centrifuge is used, but a combination of solids ejecting centrifuge followed by a liquid separator is also usable.

The sludge phase (9) is washed (operation V) with water (h) in order to increase the yield of hydrolyzate. This washing process may be performed as described for operation I. The washed phase (10) (product A) may be further enzyme treated in the same manner as phase (1) or it may be used as animal feed or as raw material for soy source or other fermented products. The wash liquid (9) is separated (operation VI) into an oil phase (11) and an oil free phase (12).

The oil phases (3), (7) and (11) from operations II, IV and VI are combined to product B from which pure soy bean oil may be isolated.

The oil-free phases (8) and (12) from operations IV and VI are combined as the raw soy protein hydrolyzate C. Product C then may be carbon treated, concentrated and dried, as described in for example United States Patent Specification No. 4,100,024.

The invention is further illustrated by the following Examples.

Example 1

600 g of full-fat soy flour (a) (Nutridan TF-100-L from Dansk Soyakagefabrik A/S) having the following composition

Protein (N x 6.25)	43.2%
	20.5%
Fat	95.0%
Dry matter	

was stepwise washed at pH 4.2. Each step includes a stirring of the solid phase and water for 30 minutes followed by a centrifugation at 3000 x g for 20 minutes in a laboratory centrifuge (Type Beckmann model J-6B). Results obtained from this washing procedure (operation I) is shown in Table 1, together with the composition of protein (N x 6.25), fat and total dry matter of the partially defatted soy flour and the combined centrifugates from the four steps. Based on these results the mass balance and yields are shown in Table 2. The word "Nutridan" is a Trade Mark, as is the word "Beckmann".

To 666.5 g of the partially defatted soy flour (1) (as sludge) which has a pH-value of 4.35 was added 39.6 ml of 4 N NaOH (f) until pH = 8.0, and 1282 g of water (d) was added to dilute the suspension to approximately 8% protein (N x 6.25). The mixture was heated to 50°C in a water bath. 3.20 g of ALCALASE 0.6 L (0.65 Anson units/g) (e) was diluted to 50 ml with water and added to the suspension containing the partially defatted soy flour (1). Thereby an enzyme activity of 13.1 Anson units per kg protein was obtained. During the hydrolysis pH was kept constant at 8.0 by addition of 4 N NaOH (f) using the pH-stat-method. The degree of hydrolysis was calculated on the bases of the consumption of base (B) by means of the relationship referred to in the reference article of J. Adler-Nissen. The DH-time relationship is shown in Figure 2. At DH = 10% 27.2 ml of 4.0 N NaOH were consumed. Then the hydrolysis was terminated by addition of DL-malic acid (g) until pH = 4.0. 44 g of DL-malic acid was used and the hydrolysis was maintained at 50°C for 30 minutes in order to inactivate the enzyme. The hydrolysis mixture was then centrifuged (operation IV) in a laboratory centrifuge (Beckmann model J-6B) at 3000 x g for 15 minutes, and 1500 g of centrifugate (6) + (7), which contains both oil and prot in hydrolyzate, and 554 g of sludge (9) was collected. The sludge phase (9) was washed with 1500 g of water (h) and centrifuged as mentioned above to yield 1500 g of centrifugates (11) + (12) and 500 g of sludge (10) (product A) (operation VI). Results obtained after performance of operations III and IV are shown in

Table 3. After having skimmed the oil phases ⑦ and ⑪ the two centrifugates ⑥ and ⑫ from operations IV and VI, were combined and adjusted to pH = 5 by use of 4 N NaOH (amount not determined) and activated carbon (BGN from Lurgi Apparate-Technik) was added in an amount of 0.2% of the total volume of hydrolyzate. After stirring for 30 minutes at 50°C the activated carbon was removed by filtration through glass fibre filter (Watman glass fibre GF/F) which has previously been washed with 5 litre of deionized water, in order to remove off-flavours from the filter. The filtrate was adjusted to pH = 6.5 and diluted to 4% protein (N x 6.25) before evaluation by means of a trained taste panel consisting of 14 persons. The hydrolyzate was compared with a sample produced from defatted flakes, as described in e.g. Fifth International Congress of Food Science & Technology, Abstracts of paper, 3b-14, "Enzymatic hydrolysis of soy protein. Processing development and applications at a low pH foods". A triangle-taste-evaluation was performed resulting in seven right answers and seven wrong answers, indicating that a taste difference could *not* be demonstrated. The word "Watman" is a Trade Mark.

TABLE 1

Centrifugate and solid phase related to operation 1.

	1. step	2. step	3. step	4. step	Combined or final
<i>Full-fat soy flour</i>					
6 N HC 1	(g) 600	-	-	-	-
Water	(ml) 34.5	0	0	0	-
	(g) 6000	6000	5000	5000	-
<i>Centrifugate:</i>					
Mass	(g) 5160	5300	5000	5000	20460
Protein conc., N x 6.25	(%) 0.25	0.07	0.07	0.04	0.10
Dry matter	(%) 2.22	0.49	0.23	0.15	0.78
Fat	(%) not determ.	not determ.	not determ.	not determ.	0.20
<i>Solid phase:</i>					
Mass	(g) 1050.6	991.4	1032.0	1009.7	1009.7
Protein conc. N x 6.25	(%) not determ.	not determ.	not determ.	23.9	23.9
Dry matter	(%) - "	- "	- "	40.7	40.7
Fat	(%) - "	- "	- "	8.2	8.2

TABLE 2

Mass balance and yields related to operation I

		Full-fat soy flour	Combined centri- fugate	Partially defatted soy flour (as sludge)	
5					5
10	Total mass (g)	600	20460	1009.7	10
	Mass of dry matter (g)	570.1	159.6	410.9	
15	Yield (%)	100	28.0	72.1	15
	Mass of pro- tein (g)	259.1	20.5	241.3	
	Yield (%)	100	7.9	93.1	
20	Mass of fat (g)	123	40.9	82.8	20
	Yield (%)	100	33.3	67.3	

TABLE 3

Results obtained after performance of operations III and IV.

	Process step and fraction	Mass of fraction, g	% Pro- tein	Yield of protein %	% Fat	Yield of fat %	
30							30
35				Based on partially defatted flour/based on full fat flour		Based on partially defatted flour/based on full fat flour	35
40	<i>Operation III</i> Partially defatted soy flour	666.5	23.9	100/93.1	8.2	100/67.3	40
45	After hydrolysis	2117.8	7.5	100/93.1	2.6	100/67.3	45
50	<i>Operation IV.</i> Centrifuge ⑦ + ⑥	1500	4.3	40.6/37.8	1.2	32.7/ 22.2	50
55	Sludge ⑧	554	not analysed		not analysed		55
	<i>Operation V</i> Centrifuge ⑪ + ⑫	1500	not analysed		not analysed		
60							60
	Product A	500	14.3	44.9/41.8	7.3	66.8/45.0	
65							65

Example 2

20 kg of full-fat soy flour (a) (Nutridan TF-100 L from Dansk Soyakagefabrik A/S) having the composition indicated in Example 1 was stepwise washed at pH = 4.2 using 4 x 180 l of water (b) of 15-20°C (operation I), acid (c) being introduced into the first step only. Each step includes a stirring of the solid phase and water followed by a centrifugation in a decanter centrifuge (Alfa-Laval N x 310-B). The sludge content in the centrifugate (determined after centrifugation of 10 ml in a graduated tube) was 2-4%. Therefore the centrifugate was re-centrifuged in a solids ejecting centrifuge Westfalia (SB 7-35-076). The results are shown in Table 4. The centrifugate indicated in Table 1 was the centrifugate from the Westfalia centrifuge and the sludge was the combined (total) sludge from the decanter and the solids ejecting centrifuge (operation I). The total combined 630 liters of centrifugates (2) were separated into 2.8 kg of an oil phase (3) and 627 kg of an oil-free phase (4) using a Westfalia centrifuge of type LG 205-2. The results obtained are shown in Table 5. The word "Westfalia" is a Trade Mark.

Based on these results the mass balance and yield related to operations I and II are shown in Table -6.

To 41 kg of the partially defatted soy flour (1) was added 46 kg of water (d) to dilute the sludge to about 6.75% protein. 685 ml of 4.8 N NaOH was added to adjust the pH to 8.0. The mixture was stirred and heated to 55°C in a tank with heating mantle. 118 g of Alcalase 0.6 L (0.65 Anson units/g) (e) was diluted to 5 liters with cold water and added to the suspension. During the hydrolysis, pH was kept constant at 8.0 by addition of 4.8 N NaOH (f) using the pH-stat-technique. A DH of 10% was reached after 133 minutes when 843 ml of 4.8 N NaOH has been consumed. Immediately thereafter, 1887 g DL-malic acid (g) was added to give a pH of 4.0.

The suspension was kept stirred at 30 minutes in order to inactivate the enzyme (operation III).

The hydrolysis mixture was then centrifuged in the solids-ejecting centrifuge (Westfalia SB 7-35-076) and 37 l of centrifugate (6) + (7) was recovered together with 50 l of diluted sludge (8). The centrifugate was then separated into 84 g of oil (7) and 34 litres of oil-free phase (6) (operation IV).

The sludge (8) was washed with 70 litres of water (h) (operation V) and separated into 73 litres of sludge (10) and 45 litres of wash liquid (9) which was separated into 43 litres of oil-free phase (12) and 66 g of oil (11) (operation VI). Results obtained during the recovery of soy protein hydrolyzate are shown in Table 7.

The oil-free hydrolyzates (6) and (12) were combined (product C), filtered, carbon treated, concentrated by reverse osmosis and freeze dried.

The oil-phases (7) and (11) were combined with the oil phase (3) from operation II, giving rise to product B.

The composition and yields of the combined products A, B and C are shown in Table 8.

It appears from the following Tables that the accuracy of the mass balances is not complete. This is due to the inaccuracy which accompanies weighing and measuring of small amounts in relatively large equipment.

TABLE 4

Centrifugate and solid phase related to operation I

	1. step	2. step	3. step	4. step	Combined or final
Full-fat soy flour (kg)	20.0	-	-	-	-
6 N HCl (kg)	1.3	-	-	-	-
Water (kg)	180.0	180	180	180	720
<i>Centrifugate:</i>					
Mass (kg)	155	160	160	155	630
Protein (% N x 6.25)	0.38	0.13	0.13	0.13	0.25
Dry matter (%)	3.30	0.43	0.50	0.21	0.96
Fat (%)	1.23	0.18	Not determ.	0.20	0.50
<i>Solid phase:</i>					
Mass (kg)	59.3	57.4	51.8	51.5	51.5
Protein (% N x 6.25)	16.44	16.0	15.06	14.19	14.19
Dry matter (%)	27.51	27.7	22.95	21.77	21.77
Fat (%)	4.12	not determ.	not determ.	1.80	1.80

TABLE 5

Results obtained after performance of operation II

	Combined centrifugate ②	Oil phase ③	Oil-free phase ④
Mass (kg)	630	2.8	627
Protein (% N x 6.25)	0.25	1.44	0.19
Dry matter (%)	0.96	62.4	0.77
Fat (%)	0.50	59.6	not determ.

TABLE 6

Mass balance and yields related to operations I and II.

	Operation I		Operation II		Oil-phase ③	Oil free phase④
	Full-fat soy flour (a)	wash liquid ②	Partially de- fatted soy flour①			
Total mass	(kg)	630	51.5	2.8	627	
Mass of DM	(kg)	6.05	11.2	1.75	4.83	
Yield	(%)	31.8	59.0	9.2	25.4	
Mass of protein	(kg)	1.58	7.31	0.04	1.19	
Yield	(%)	18.5	84.6	0.5	13.9	
Mass of fat	(kg)	3.15	0.92	1.67	not determ.	
Yield	(%)	76.8	22.4	40.7		

TABLE 7
Results obtained after performance of operations III, IV, V and VI

Operation and fraction	Mass of fraction kg	% protein	Yield of protein % based on partially defatted flour	% fat	Yield of fat based on partially defatted flour	Yield of fat based on full-fat flour
Operation III						
①	41	14.19	100	1.80	100	22.4
⑤	95.5	6.56	—	(1.87)	—	—
Operation IV						
⑦	0.084	2.63	0.04	60.4	6.9	1.5
⑧	34	4.38	25.6	—	—	—
③	50	7.63	65.6	not analyzed	—	—
Operation V						
⑩ A	73	2.75	34.5	not analyzed	—	—
⑨	45	1.88	14.5	not analyzed	—	—
Operation VI						
⑪	0.066	2.53	0.03	61.5	5.5	1.2
⑫	43	1.81	13.4	—	—	—

() means that the figures is unrealistic.

TABLE 8

Composition and yields of product A, B and C based on full-fat soy flour

5	Compon nt	A	B	C	5
	Protein %	2.75	1.5	2.99	
	Yield %	29.2	0.5	33.0	
10	Dry matter %	10.3	65	4.5	10
	Yield %	(84.2)	10	22.9	
15	Oil %	not determ.	60	—	15
	Yield %	not determ.	43.4	—	

() means that the figure is unrealistic.

20 CLAIMS

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1. A method of producing soy protein hydrolyzate from fat-containing soy material (as defined), which method comprises hydrolyzing a partially defatted solid soy material, obtained by washing a fat-containing soy material in an aqueous medium at a pH in the range of from 3.5 to 5.5 at a relatively constant pH with a proteolytic enzyme in the presence of water and a base to a DH in the range of from 1 to 20 and thereafter deactivating the enzyme, whereafter the aqueous hydrolyzate phase is separated from the oil phase and the solid phase.
2. A method according to Claim 1, which includes the step of washing fat-containing soy material in an aqueous medium having a pH in the range of from 3.5 to 5.5.
3. A method according to Claim 2, wherein the step of washing the fat-containing soy material is washed in an aqueous medium having a pH in the range of from 4.2 to 4.5.
4. A method for production of soy protein hydrolyzate from fat-containing soy material, wherein the fat-containing soy material (a) is washed in an aqueous medium at pH 3.5 to 5.5 (operation I), preferably from 4.2 to 4.5, whereby the wash water (2) from operation I is introduced into a separator, wherein it is separated into an oil phase (3) and a waste water phase (4) (operation II), and whereby the washed, partially defatted, solid soy material (1) from operation I is introduced into a hydrolysis container, to which also water (d), a proteolytic enzyme (e) and base (f) is added, in which hydrolysis container the partially defatted soy material (1) from operation I is hydrolyzed at a relatively constant pH to a DH of between 1 and 20 (operation III), whereafter the proteolytic activity is inactivated, whereby the slurry (5) from operation III is introduced into a separator, in which the slurry is separated into an oil phase (7), an aqueous hydrolyzate phase (6) and a sludge phase (8) (operation IV), whereby the sludge phase (8) from operation IV is collected (product A), whereby the oil phases (3) and (7) from operations II and IV are combined (product B) and whereby the aqueous hydrolyzate phase (6) from operation IV is collected (product C).
5. A method according to Claim 4, wherein the sludge phase (8) from operation IV before collection is transported to a washing device, to which also water (h) is added (operation V), whereafter the precipitate (10) from operation V is collected as product A, the wash water phase (9) from operation V is introduced into a separator, in which it is separated into an oil phase (11) and an aqueous hydrolyzate phase (12) (operation VI), the oil phases (3), (7) and (11) from operations II, IV and VI are combined (product B) and the aqueous hydrolyzate phases (6) and (12) from operations IV and VI are combined (product C).
6. A method according to Claim 4 or 5, wherein the separations in operations II and IV or II, IV and VI are performed by means of a centrifuge.
7. A method according to Claim 1 or 6, wherein the proteolytic enzyme (e) used for the hydrolysis is produced by means of *B. licheniformis* and the hydrolysis (operation III) is performed around the pH optimum of this enzyme.
8. A method according to Claim 1 to 7, wherein the hydrolysis (operation III) is performed at a pH which does not differ more than 2.5 pH units from the optimum pH of the proteolytic enzymes.
9. A method according to Claim 1 to 8, wherein the hydrolysis (operation III) is carried out to a DH in the range of from 8 to 12.
10. A method according to Claim 1 to 9, wherein the proteolytic activity is inactivated by means of malic or citric acid.
11. A method of producing soy protein hydrolyzate substantially as hereinbefore described with reference to Figure 1 of the accompanying drawings.
12. A method of producing soy protein hydrolyzate substantially as hereinbefore described with reference to Figure 2 of the accompanying drawings.
13. A method of producing soy protein hydrolyzate substantially as described in any one of the foregoing

Examples.

14. A soy prot in hydrolyzate whenever produced by the method of any one of the preceding claims.
15. Any novel featur or combination of features described herein.

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